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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.
09/276,820	03/26/99	HARRINGTON J	1522.0030004

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EXAMINER	
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ART UNIT	**PAPER NUMBER**
1632	19

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Please find below and/or attached an Office communication concerning this application or proceeding.

Commissioner of Patents and Trademarks

Office Action Summary

Application No.	Applicant(s)
09/276,820	Harrington et al
Examiner	**Group Art Unit**
Ram Shukla	1632

☒ Responsive to communication(s) filed on *May 4, 2000*

☐ This action is **FINAL.**

☐ Since this application is in condition for allowance except for formal matters, **prosecution as to the merits is clos d** in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11; 453 O.G. 213.

A shortened statutory period for response to this action is set to expire *three* month(s), or thirty days, whichever is longer, from the mailing date of this communication. Failure to respond within the period for response will cause the application to become abandoned. (35 U.S.C. § 133). Extensions of time may be obtained under the provisions of 37 CFR 1.136(a).

Disposition of Claim

☒ Claim(s) 58,59,64-69,71-74,76-82,85-123,128,129,131-132,157,159,161,162,164-167,169-175,177-183,223-226,232-259 is/are pending in the applicat

Of the above, claim(s) _____ is/are withdrawn from consideration

☐ Claim(s) _____ is/are allowed.

☒ Claim(s) 58,59,64-69,71-74,76-82,85-89,98,105,106,109-123,129,132,157,159,161,162,164-167,169-175,177-183,223-226,232-259 is/are rejected.

☒ Claim(s) 90-97,99-104,107,108,128 and 131 is/are objected to.

☐ Claims _____ are subject to restriction or election requirement.

Application Papers

☐ See the attached Notice of Draftsperson's Patent Drawing Review, PTO-948.

☐ The drawing(s) filed on _____ is/are objected to by the Examiner.

☐ The proposed drawing correction, filed on _____ is ☐ approved ☐ disapproved.

☐ The specification is objected to by the Examiner.

☐ The oath or declaration is objected to by the Examiner.

Priority under 35 U.S.C. § 119

☐ Acknowledgement is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d).

☐ All ☐ Some* ☒ None of the CERTIFIED copies of the priority documents have been

☐ received.

☐ received in Application No. (Series Code/Serial Number) _____.

☐ received in this national stage application from the International Bureau (PCT Rule 17.2(a)).

*Certified copies not received: _____

☐ Acknowledgement is made of a claim for domestic priority under 35 U.S.C. § 119(e).

Attachment(s)

☒ Notice of References Cited, PTO-892

☒ Information Disclosure Statement(s), PTO-1449, Paper No(s). *12*

☐ Interview Summary, PTO-413

☐ Notice of Draftsperson's Patent Drawing Review, PTO-948

☐ Notice of Informal Patent Application, PTO-152

--- SEE OFFICE ACTION ON THE FOLLOWING PAGES ---

DETAILED ACTION

1. Amendment filed 05-04-200 (paper #14) has been entered. Supplemental amendment filed 7-14-00 (paper #17) has been entered

2. Claims 1-7, 10-15, 20-36, 60-63, 70, 75, 83, 84, 94, 95, 124-127, 130, 133-156, 158, 160, 163, 168, 176, 184-222, and 227-231 have been canceled.

3. Amended claims 64, 69, 76, 77, 81, 89, 99, 104, 107, 108, 118, 128, 129, 159, 161, 162, 165, 166, 173, 174, 180, and 181 have been entered.

4. New claims 232-259 have been entered.

5. Claims 58, 59, 64-69, 71, 73-74, 76-82, 85-123, 128, 129, 131, 132, 157, 159, 161, 162,164, 165-167, 169-175, 177-183, 223-226, 232-259 are pending in the instant application.

Priority

6. Applicant has not complied with one or more conditions for receiving the benefit of an earlier filing date under 35 U.S.C. 112 as follows:

> The second application (which is called a continuing application) must be an application for a patent for an invention which is also disclosed in the first application (the parent or provisional application); the disclosure of the invention in the parent application and in the continuing application must be sufficient to comply with the requirements of the first paragraph of 35 U.S.C. 112. See *In re Ahlbrecht*, 168 USPQ 293 (CCPA 1971).

In the instant case, the specification as filed does not disclose the limitations of the claims 235-259, therefore, these claims do not meet the condition for receiving the benefit of an earlier filing date. Accordingly, the date on which claims 235-259 were entered in the application will be considered as the effective filing date for these claims, i.e. 7-14-00.

7. Declaration Under 37 CFR 1.132, filed 5-4-00 has been entered. However, in the light of the new non-final rejection and new grounds of rejection, Applicants declaration and Arguments in response to previous office action of 2-1-00 are moot.

Claim Objections

8. Claims 90-97, 99-104, 107, 108, 128, and 131 are objected to under 37 CFR 1.75(c) as being in improper form because a multiple dependent claim can not depend from another multiple dependent claims. See MPEP § 608.01(n). Accordingly, claims 90-97, 99-104, 107, 108, 114, 115, 128, and 131 have not been further treated on the merits.

9. Claim 259 is objected to under 37 CFR 1.75(c), as being of improper dependent form for failing to further limit the subject matter of a previous claim. Applicant is required to cancel the claim(s), or amend the claim(s) to place the claim(s) in proper dependent form, or rewrite the claim(s) in independent form.

Claim 259 limits the method of claim 254 by reciting that the vector is a retrovirus vector construct. However, claim 254 itself depends on claim 242 which recites a retroviral vector construct. Therefore, it is not clear how claim 259 is further limiting the method of claim 254.

Double Patenting

10. A rejection based on double patenting of the "same invention" type finds its support in the language of 35 U.S.C. 101 which states that "whoever invents or discovers any new and useful process ... may obtain a patent therefor ..." (Emphasis added). Thus, the term "same invention," in this context, means an invention drawn to identical subject matter. See *Miller v. Eagle Mfg. Co.*, 151 U.S. 186 (1894); *In re Ockert*, 245 F.2d 467, 114 USPQ 330 (CCPA 1957); and *In re Vogel*, 422 F.2d 438, 164 USPQ 619 (CCPA 1970).

A statutory type (35 U.S.C. 101) double patenting rejection can be overcome by canceling or amending the conflicting claims so they are no longer coextensive in scope. The filing of a terminal disclaimer cannot overcome a double patenting rejection based upon 35 U.S.C. 101.

11. Claims 234-259 provisionally rejected under 35 U.S.C. 101 as claiming **the same invention** as that of claims 58-60, 64--69 of copending Application No. 09/515,123, filed 1-27-2000. This is a provisional double patenting rejection since the conflicting claims have not in fact been patented.

It is noted that claims 244, 246, 251, 253,255,257 of the instant application are the same as the claims 58, 59, 60, 64, 65, and 68 of the the co-pending Application 09/515,123.

Claim Rejections - 35 U.S.C. § 112

12. The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

13. Claims 235-259 are rejected under 35 U.S.C. 112, first paragraph, as containing subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention.

An objective standard for determining compliance with the written description requirement is, "does the description clearly allow persons of ordinary skill in the art to recognize that he or she invented what is claimed." To satisfy the written description requirement, an applicant must convey with reasonable clarity to those skilled in the art that, as of the filing date sought, he or she was in possession of the invention, and that the invention, in that context, is whatever is now claimed. The test for sufficiency of support in a parent application is whether the disclosure of the application relied upon "reasonably conveys to the artisan that the inventor had possession at that time of the later claimed subject matter." (MPEP 2163).

In the instant case, Claim 234 contains a negative limitation that the vector does not contain a polyA site operably linked to the transcriptional regulatory sequence, however, nowhere in the specification, such a limitation of the transcriptional regulatory sequence has been disclosed. Claims 235-259 contain negative limitations that describe the constructs and methods using such constructs, for example, in these claims, the exon is recited as not to be an antibiotic resistance activity, or is not a reporter gene or wherein there is no selectable marker between the transcriptional regulatory sequence and the splice donor sequence or wherein said exon does not contain a selectable marker, there is no ribosomal entry site between the transcriptional regulatory sequence and the splice donor sequence, the exon does not contain an internal ribosomal entry site. However, as filed the specification does not describe any of these definitions of the exon of the claimed vectors. Likewise the limitations that the exon or the splice donor sequence is derived from a naturally occurring eukaryotic splice donor sequence is also not disclosed in the specification and the applicant had not pointed out where these limitations have been disclosed in the specification and persons skilled in the art at the time the application

was filed would not have recognized the description of these limitation in the disclosure of the application as filed.

Accordingly, it is concluded that the written description requirement is not satisfied for the claims 234-259 and the subject matter of claims 234-259 is considered new matter.

14. Claims 58, 59, 64-69, 71, 76-80, 85-89, 98, 105, 106, 109-123, 129, 157, 180-183, 223-226, 232-259 are rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for a vector wherein the vector sequence elements are linked in a certain order, for practicing claimed methods in isolated eukaryotic cells in vitro, does not reasonably provide enablement for any other embodiments. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the invention commensurate in scope with these claims.

For the vector of claims 58, the order of the sequence elements operably linked in the 5' to 3' direction is a promoter, a nucleic sequence encoding an amplifiable marker and an unpaired splice donor sequence. For the vector of claim 59, the order of the sequence elements operably linked in the 5' to 3' direction is a viral origin of replication, a promoter and a nucleic acid sequence encoding an amplifiable marker. For the vector of claim 157, the order of the sequence elements operably linked in the 5' to 3' direction is a first promoter, a nucleic acid sequence encoding a positive selection marker, an unpaired splice donor site, a second promoter, and a nucleic acid sequences encoding a negative selectable marker.

While determining whether a specification is enabling, one considers whether the claimed invention provides sufficient guidance to make and use the claimed invention, if not, whether an artisan would require undue experimentation to make and use the claimed invention and whether working examples have been provided. The specification is not enabling for the claimed invention because the specification does not provide sufficient guidance, evidence or exemplification so that an artisan of skill would have been able to make and use the invention as claimed invention without undue experimentation.

As summarized previously (paper # 11), instantly claimed methods are drawn to activation of any endogenous gene or to a list of over 70 genes using a collection of vectors that may comprise only a transcriptional regulatory sequence alone or in combination with splice donor site and one or more selectable markers and one or more promoters. The claimed invention is different from a method of targeted gene activation in that it does not use a targeting sequence that will have homology to a known gene, in other words integration of the vectors are not by homologous method but by random integration and non-homologous recombination. The specification has provided, diagrams of vectors that comprise claimed sequence elements and protocols to make libraries, PCR and other techniques of making cell lines, library etc.

First issue is: will the vectors work for intended use if different sequence components of the vector (such as the promoter, exon, splice donor site etc.) are present in any order and comprising any other sequence elements because all the vector claims recite a vector comprising. As an example, the intended use of the claimed invention is to join the unpaired spliced donor site of the vector with a splice acceptor site of an endogenous gene so that the promoter of the vector can drive the expression of the endogenous gene and by screening the expression of the marker gene cells which express such gene can be identified. For example, in claim 58 if the amplifiable marker was present 3' to the splice donor site, once integrated into the genome of a cell, it would be spliced out as an intron when the splice donor site is spliced with a splice acceptor site of an endogenous gene, accordingly, the method would not work. A similar case can be made for the presence of the sequence elements of all other vectors or methods using such claims. As another example, lets consider claim 157, the claimed vector would work for the intended use if the sequence elements are in the order of a, b and c. If one had to put the splice donor site first, all the marker genes would be spliced out. There can be other combinations of these sequence elements which again would not result in the vector usable for its intended use.

Next, will the vectors work for intended use if the transcriptional regulatory sequence was not a promoter, but it was only an enhancer? The specification as filed does not provide sufficient guidance as to whether a vector comprising an enhancer linked to a spliced donor site or other sequence elements would have activated the expression of endogenous genes. For

example, if one integrated a vector comprising a CMV immediate early enhancer without promoter linked to splice donor site or amplifiable marker or both in the genome of a cell after the native promoter of an endogenous gene, the claimed vector would not have induced the expression of the endogenous gene because such an enhancer would have not initiated transcription of the gene. For initiating transcription of a sequence, a promoter should have binding sites wherein basic transcription factors can bind and cause the formation of a transcription initiation complex. If the enhancer containing vector was to be incorporate upstream of a promoter of an endogenous gene it may result in the deletion of the promoter of the endogenous gene due to the pairing of the unspliced sequences, if transcription could be initiated from the enhancer (???). Therefore, an artisan would not have been able to use the claimed vectors for the intended use if a transcriptional regulatory sequence other than promoter was used.

Next, the question is: will claimed vectors and methods work in any and all cells? For example, these vectors would not work in prokaryotic cells which do not have introns in their genes or which do not have splicing factors or splicing machinery. Likewise the retroviral vectors or other viral vectors and promoters would not work in prokaryotic cells because these cells would be lacking in transcription factors that would be needed for the LTRs and eukaryotic promoters to initiate and/or activate transcription. It is noted that these are basic facts of transcriptional regulation well known in the art.

Next, in regard to claims 77, 79, 80, 86-88, 98, 109-117, 223, 226, and 232 the issue is: will the methods work in vivo? Activation or over-expression of a gene in vivo or by providing cells, in which the expression of an endogenous gene has been activated in vitro, to animals is highly unpredictable because of a multitude of factors. At the time of the invention, both in vivo gene therapy method as well as ex-vivo gene therapy methods were unpredictable.

Crystal (Crystal RG. Science 270:404-410.1995) in assessing the state of the art of the gene therapy noted that gene therapy still faced significant hurdles such as in the areas of inconsistent results, extrapolation of studies from experimental animals to humans, vector etc. He also adds that there are several examples wherein prediction of gene transfer studies in experimental animals have not be borne out in human trials (see para 4 in col 1 on page 409).

Then there is the issue of production of vectors, free of aggregation, contamination and variability from preparation to preparation, some of the problems that must be overcome before large clinical trials can be initiated. Additionally, there is the issue of an ideal vector is conceptually impractical because the human applications of gene transfer are broad and the ideal vector will likely be different for each application (see col 2 on page 409).

In a more recent assessment of the gene therapy art, Verma and Somia (Verma IM and Somia N. Nature 389: 239-242. 1997) summarize " But the problems- such as lack of efficient delivery systems, lack of sustained expression, and host immune response reactions-remain formidable challenges" (see the abstract). Although more than 200 clinical trials are currently underway worldwide, with hundreds of patients enrolled, there is no single outcome that we can point to as a success story" (see first and second paragraphs in col 1 on page 239).

Anderson (Anderson WF. Nature 392 (SUPP):25-30, 1998) notes that since the approval of first clinical trial of gene therapy protocol in 1990, more than 300 protocols have been approved worldwide. He further adds, "The conclusions from these trials are that gene therapy has the potential for treating a broad array of human diseases and that the procedure appears to carry a very low risk of adverse reactions; the efficiency of gene transfer and expression in human patients is, however, still disappointingly low. Except for anecdotal reports of individual patients being helped, there is still no conclusive evidence that a gene therapy protocol has been successful in the treatment of a human disease."

Finally, the method of ex vivo gene therapy wherein a cell is altered first by introducing a DNA of interest into the cells and then the cells are administered to a subject is unpredictable. According to Kay et al. (Kay MA et al. Proc. Natl. Acad Sci. 94: 12744-12746, 1997), this method is very inefficient. For example, in the most commonly used method of ex vivo gene therapy wherein hematopoietic cells are altered in vitro, only small percent of cells (less than 3%) are transduced (see column 1 on page 12746). It is not clear question becomes whether such a low percent of transduced cells would be able to provide the desired levels of gene expression in the

instant case where the method has a shortcoming that it may not produce a full length or biologically active protein.

In addition to the above discussed issues of general nature related to in vivo and ex vivo gene therapy, in the instant case, it is not certain that the methods would have produced a full length protein or an active protein and if the protein is inactive or is a fraction, will the method work as intended. Even if one had to think that the in vivo methods are for producing antibodies if the method is not certain whether a full length protein is produced or a fragment is produced and whether such fragments are antigenic and whether there will be any antibody production, how would an artisan know how to use the claimed invention of the vectors and methods in vivo. It is noted that the integration of the vector in the genome will be random in the claimed method, however, it is not clear as to how a given gene would be activated in vivo when the method is to be carried out in an animal or a human (claims 131 and 132) because the vector may get introduced primarily in cells at site of the administration and may not even reach the target cells. For example, if the method is to increase the expression of human growth hormone in an animal or a human, there is nothing in the method to ensure that the animal or human would have activated a growth hormone gene or an oncogene or a tumor suppressor gene, if not how would an artisan know how to activate only growth hormone gene in a human or an animal in vivo using the claimed methods.

Regarding the claims 104-106, it is noted that an artisan would not know whether the vector construct of the claimed method would integrate at the site of the double strand break because as emphasized by the specification, the method works due to random integration. There is nothing in the method steps or in the specification which would ensure that the vector construct of the claimed invention would have integrated at the site of DNA breaks. Therefore, an artisan would not even know whether the steps of DNA break would have any effect on the practicing of the method as recited.

In conclusion, the claimed invention is not enabled because the specification as filed does not provide sufficient guidance and/ working examples as to how an artisan of skill would have made and practiced the claimed invention commensurate in scope with the claims and

therefore, limiting the scope of claimed invention to vectors that have the sequence elements in the order recited and wherein the cells are isolated eukaryotic cells and the method is carried out in vitro, is proper.

15. The following is a quotation of the second paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

16. Claims 72, 73, 77-82, 85-89,98, 105--106, 109-123, 129, 157, 159, 161, 162, 162, 165 - 167, 169-175, 177-183, 223-226, 232-259 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Claim 72 is incomplete because it is dependent on a canceled claim. Accordingly claim 72 and its dependent claims could not be examined.

Claim 73 is indefinite because it is dependent on an indefinite claim, claim 72.

Claims 77 and 81 are indefinite because the term "the construct" does not have an antecedent basis and because the base claim recites "a vector construct", not "a construct."

Claims 77 and 113 are indefinite because the term "said endogenous gene" does not have an antecedent basis and because the preamble of the claim recites the term "an endogenous cellular gene."

Claim 81 is also indefinite because the term "said construct" in line 2 does not have an antecedent basis.

Claims 85-87, 109-110, 116 are indefinite because the term "said gene" does not have an antecedent basis.

Claim 89 is also indefinite because it is dependent from claim 232 which is not a preceding claim.

Claims 118 and 119 are indefinite because the term "genetic construct" does not have an antecedent basis.

Claim 157 is indefinite because the term "said splice donor site" does not have an antecedent basis.

Claims 159, 161, 162, 165-167, 169-171, 174, 175, and 177-179 are indefinite because the term "said vector or the vector" does not have an antecedent basis.

Claim 180 is indefinite because it is unclear whether the term "an endogenous gene" is same in the preamble and the body of the claim.

Claim 181 is indefinite because the terms "the host cell", "said cells", and "said selected cells" do not have an antecedent basis.

Claim 223 is vague and indefinite because it is unclear as to what would be considered "suitable for activating an endogenous gene."

Claim 232 is indefinite because it is unclear as to whether an endogenous gene recited in the preamble of the claim is same as an endogenous gene recited in the body of the claim (c).

Claim 232 is indefinite because the term "said isolated and cloned cell" does not have an antecedent basis.

Claim 224 is indefinite because the term "said host cell" does not have an antecedent basis.

Claim 234 is vague and indefinite because it is incomplete and does not have a period at the end.

Claims 234-241, 243-245, 247, and 257 are indefinite because the term "said construct" does not have antecedent basis.

Claim 236 is vague and indefinite because it recites the term "said exon does not contain a selectable marker." A selectable marker is a protein while an exon is a DNA sequence, how can a DNA sequence contain or not contain a protein sequence.

Claims 58, 59, 157 are vague and indefinite because the recite " a vector construct comprising.....amplifiable marker....." or a vector construct comprising.....selectable marker.....". A marker (selectable or amplifiable) is a protein while a vector construct is a nucleic acid sequence, how can a nucleic acid sequence contain or not contain a protein sequence.

Claim 246 is indefinite because the term "the vector construct" does not have an antecedent basis.

Claims 249-250 are indefinite because the term "the construct" does not have an antecedent basis.

Claims 256, 257, and 258 are indefinite because the term "said cell" does not have an antecedent basis.

Claim 257 is indefinite because the term "said eukaryotic cell" does not have an antecedent basis.

Claim 159 is vague because it is unclear as to what is meant by "a transposition signals."

Claim Rejections - 35 U.S.C. § 102

17. The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

> A person shall be entitled to a patent unless --
>
> (b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

18. Claims 58, 64, 69, 74, 76, 234-241, 243, and 247 are rejected under 35 U.S.C. 102(b) as being anticipated by Reilly et al. (Reilly JD et al. DNA and Cell Biology 9:535-542, 1990).

Reilly et al teach transcription vectors for identification and mapping of RNA splice sites in genomic DNA (see figure 1B) wherein the vector has a T7 promoter linked to Exon 1 of human growth hormone and some intronic sequences. It is noted that the vector is based on the bluescript phagemid of Stratagene pBS which contains LacZ gene which is amplified since pBS is a multicopy plasmid. Since the vectors and methods of Reilly would involve transformation of cells with the plasmid to amplify the plasmid, cells and host cells comprising the vector are inherent to the method.

Accordingly, the invention of claims 58, 64, 69, 74, 76, 234-241, 243, and 247 is anticipated by Reilly et al.

19. Claims 59, 69, 71, 73, 74, 76, 225, and 226 are rejected under 35 U.S.C. 102(e) as being anticipated by Treco et al (US 5,641,670 dated 01-24-1997).

Treco et al teach vectors for protein production and delivery, for example, the vector of figure 8 which has a transcription regulatory sequence, hGH exon 1, dhfr gene and origin of replication. This patent also teaches a cell wherein the vector is integrated, and wherein an endogenous gene is over-expressed in said cell by up regulation of the gene by transcriptional regulatory sequence on said vector construct (see figures, examples and columns 1, lines 56-67 continued in columns 6 for summary).

Accordingly, the invention of claims 59, 69, 71, 73, 74, 76, 225, and 226 is anticipated by Treco et al.

20. Claims 77, 78, 80-82, 87-89, 109, 114, 116-123, 129, 157, 159, 161, 162, 164, 165, 166, 167, 169-175, 177-183, 223, 224. 232, and 233 are rejected under 35 U.S.C. 102(b) as anticipated by or, in the alternative, under 35 U.S.C. 103(a) as obvious over Treco et al as evidenced by Capecchi et al (Capecchi MR. Scientific American , March 1994, pages 52-59).

Some of the teachings of Treco et al have been summarized above (paragraph 19). It is noted that in figures 1-14, Treco et al have disclosed the structure of their constructs and their sequence elements. Treco et al also teach examples to increase the production of proteins encoded by genes present in a cells genome, such as the expression of erythropoietin , human growth hormone, screening of cells expressing the gene product (see examples 1-5).

Capecchi reviews the state of the art of gene replacement by homologous recombination. Capecchi notes, "Regrettably, such targeted replacement occurs only in a small fraction of the treated cells. More often, the targeting vector inserts randomly at non-matching sites or fails to integrate at all. We must therefore sort through the cells to identify those in which targeting has succeeded. Approximately, one in a million treated cells has the desired replacement" (see last paragraph in column 3 on page 56 continued in the first paragraph in column on page 57). He further discusses the methods to screen the cells which express the targeted gene.

It is noted that the vector components are same in the Treco method and the claimed method because the vectors and methods of the claimed method use the term "comprising" which would indicate that any other sequence element can be present. Although, the claimed

method of the instant application recites in step b (claim 77) 'integrating said construct into the genome of said cell by non-homologous recombination', however, as noted by Capecchi, even when homologous sequences are included in a vector for targeted activation of an endogenous gene, the vector integrates in the genome of the majority of the cells by non-homologous recombination because that is the preferred method of integration of an exogenous DNA in a cell. While the method of Treco is to homologous recombination method, it would include majority of cells wherein the vector would have integrated randomly and then a cell with proper expression product has to be screened which would be the same as claimed in the instant application.

In the event that the claimed protein is not identical to that disclosed by Treco, it is considered that, as noted above, integration of the Treco et al construct will occur by random integration and if integrated 3' to an endogenous promoter would expression the coding sequences downstream of the site of integration. Thus the claimed invention as a whole was at least obvious over, if not anticipated by, the prior art.

Therefore, the invention of claims 77, 78, 80-82, 87-89, 109, 114, 116-123, 129, 157, 159, 161, 162, 164, 165, 166, 167, 169-175, 177-183, 223, 224. 232, and 233 is anticipated by Treco et al.

21. Claims 109-113, 115 are rejected under 35 U.S.C. 102(b) as anticipated by or, in the alternative, under 35 U.S.C. 103(a) as obvious over by Joshi (Joshi S. Medical Hypothesis. 36: 242-245, 1991).

Joshi teaches vectors and methods for activating expression of genes which are silent or poorly expressed by randomly integrating retroviral vectors. In figure 1, Joshi provides a flow chart for the method. This prior art also teaches that cells expressing a marker gene can be selected. Alternatively, cells expressing a particular gene activated by the method can be screened by using an assay that is based on the gene, for example, ligand binding assay (see second paragraph on page 244). Joshi further teaches that once a mutant expressing the desired phenotype is screened it could be used for further applications (e.g. enzyme kinetics, drug testing) as well as genomic or cDNA cloning of the insertionally activated gene responsible

for the phenotype (see paragraph 2 on page 244). Joshi also teaches the structure of the vectors wherein gag, pol and env genes have been deleted but the vector has a 5' sequence which has the packaging signal and splice donor site (see Figure 2 and description in paragraph 1 on page 244).

It is noted that the most important step of the claimed method is the vector and its integration into cellular genome randomly, which is the main teaching of Joshi. Other steps are art recognized conventional steps of cells culture and protein screening. Therefore, the invention of claims 109-115 is anticipated by Joshi.

In the event that the claimed protein is not identical to that disclosed by Joshi, it is considered that any differences would be the result of minor variations, for example, a method of screening for gene expression, candidate compounds as drugs, cell culture, such variations would have been obvious over the prior art. Thus the claimed invention as a whole was at least obvious over, if not anticipated by, the prior art.

22. Claims 59, 69, 74, 76, 225 and 226 are rejected under 35 U.S.C. 102(b) as being anticipated by Benedetti et al (Nucleic Acid Research 19: 1925-1931, 1991).

Benedetti et al teach a BK virus-based episomal vector for expression of foreign genes in mammalian cells. Figure 1 teaches the structure of the vector which comprises inducible MMTV and viral origin of replication. MMTV promoter is an inducible promoter that would result in induced expression of the gene comprised in the vector (see the abstract). Benedetti et al note that the vector replicates to high copy number and the copy number can be varied by changing the G418 concentration in the culture medium.

Accordingly, the invention of claims 59, 69, 74, 76, 225 and 226 is anticipated by Benedetti et al.

23. Claims 234-259 are rejected under 35 U.S.C. 102(b) as being anticipated by Zambrowicz et al (Nature 392:608-611, 1998).

Zambrowicz et at teach disruption, expression analysis and sequence identification of 2,000 genes in mouse embryonic stem cells, using a retroviral vector wherein the vector comprises a first LTR, a promoter, a marker gene, an unpaired spliced donor site and a second LTR (see the abstract and Figure 1). They also disclose activation of genes as evidenced by expression analysis by RT-PCR method (see Figure 4).

Accordingly, the invention of claims 234-259 is anticipated by Zambrowicz et al.

24. Claims 234-259 are rejected under 35 U.S.C. 102(e) as being anticipated by Zambrowicz et al (US 6,080,576, 6-27-00).

Zambrowicz et al teach a vector comprising a marker gene, operably linked to a promoter, a second promoter linked to an exon sequence such that the exon is 3' to the second promoter and the second promoter expresses the exon and wherein the exon at its 3' end is defined with a splice donor sequence and the exon and the splice donor sequences are derived from a naturally occurring gene. Zambrowicz et al teach a retroviral vector that comprises a retroviral LTR, a promoter operably linked to an exon which at its 3' end has splice donor sequence and second retroviral LTR. Zambrowicz et al teach that the exon could not be a reporter gene or could not be a selectable marker gene or antibiotic resistance gene (see claims 5-15). Zambrowicz et al also teach method of trapping endogenous genes and activating endogenous genes, making library of cells wherein the vector is inserted into the genome of the cells, identifying genes that are induced by signals or phenotype of interest. Examples in columns 28-32 teach the method of the construction of the vectors and gene trapping.

Accordingly, the invention of claims 234-259 is anticipated by Zambrowicz et al.

Claim Rejections - 35 U.S.C. § 103

25. The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

26. Claims 65-68 and 71 are rejected under 35 U.S.C. 103(a) as being unpatentable over Reilly et al. in view of Kaufman 1995 (Short Protocols in Molecular Biology, ed. Ausubel et al. John Wiley & Sons, Inc. 1995, pages 16-58-16-60) and Dominski et al. (Mol. Cell. Biol. 11:6075-6083, 1991).

Claims 65-68 limit the vector construct of claim 64 wherein the promoter is a viral promoter, a CMV promoter, a non-viral promoter, and an inducible promoter. Claim 71 recites that the cell of claim 69 is integrated into the cellular genome.

Teachings of Reilly et al has been summarized previously (paragraph 15). Reilly et al do not teach that the vector in their construct is a viral promoter, a CMV promoter, a non-viral promoter, and an inducible promoter.

Kaufman reviews strategies for expressing foreign genes into mammalian cells, both for transient expression as well as stable expression. This prior art compares the expression levels from different promoters and expression systems, their use in transient or stable expression and primary uses. Kaufman further teaches that is a selection procedure is applied after DNA transfection, it is possible to isolate cells that have stably integrated foreign DNA into their genome. This prior art teaches all the methods used for isolating stably transfected cells (see entire article).

Dominski et al. disclose role of exon length in splice site selection for pre-mRNA processing. They further teach to sub-clone the constructs for in vitro splicing in plasmids that have CMV promoter for in vivo studies (see materials and methods section, page 6076, lines 1-11).

At the time of the invention it would have been obvious to one of ordinary skill in the art to modify the vector of Reilly et al by replacing the T7 promoter in their splicing vectors with different promoters, such as a viral vector or an inducible promoter as taught by Kaufman and test their splicing efficiency in vivo in eukaryotic cells. An artisan could also include a selection marker and isolate cells that were neo resistant, wherein vector DNA would have integrated in

the genome of the selected cells with reasonable expectation of success because all the relevant methods are reviewed by Kaufman and different protocols are also provided and it is reiterated that these methods, of cloning promoters and producing stable cells lines is routine in any cell biology or molecular biology laboratory. An artisan would have been motivated to use recited promoters because this would have allowed testing the splicing efficiency in vivo in a cell (as done by Dominski et al.) and a promoter of choice would be used keeping in mind the conventional wisdom that the expression level of foreign genes under the control of different promoter varies greatly based on the cell type used as discussed by Kaufman (see section on choice of expression system on page 16-60). Furthermore, an artisan would have been motivated to select cells that would integrated the vector in their genome because stable transfectants would have helped in producing cell lines that would have expressed a target coding sequence constitutively. Regarding CMV promoter, it is noted that CMV promoter is yet another commonly used promoter for expressing foreign genes in a mammalian cells (as used by Dominski et al.).

27. Claims 159 and 167 are rejected under 35 U.S.C. 103(a) as being unpatentable over Reilly et al., as applied to claims 58, 64, 69, 74, 76, 234-241, 243, and 247 above, and further in view of Devine et al (US 5,677,170, 10-14-1997) and Joshi et al.

The teachings of Reilly et al and Joshi have been summarized previously (paragrapsh 18 and 21 respectively). None of these teach to include transposition signal in their vectors.

Joshi teaches that the use of transposons in vectors is a known method of cloning new bacterial genes by producing bacterial mutants by random integration of plasmids containing transposons and then identifying and cloning the mutated genes (see last paragraph in column 2 on page 242).

Devine et al teach efficient methods of creating artificial transposons and inserting these transposons into plasmid targets in vitro. These can be used to introduce any functional or non-functional DNA cis elements, sequence or combination into another segment of DNA (see lines 11-67 col 10 continued in cols 11 and 12). They also teach that in their experiments the

transposon integration withing targets was sufficiently random that insertion could be recovered from all regions of target plasmids (see lines 17-19, column 6). Additionally, Devine et al disclose that transposons can be used for mapping genes, chromosome fragments, promoters, enhancers, introns and exons (see columns 9-11).

At the time of the invention, it would have been obvious to an artisan of ordinary skill in the art to modify the vector of Reilly et al by including transposition sequences of Devine et al with reasonable expectation of success and use such vectors in cloning new genes by the method of Joshi or identify splice sites in DNA fragments or in genomic sequences in a cell. An artisan would have been motivated to include the transposon in the vector because it would result in the integration of the vector at different sites in the genome of a cell due to random integration.

28. Claims 159 and 167 are rejected under 35 U.S.C. 103(a) as being unpatentable over Treco et al. as applied to claims 59, 69, 71, 73, 74, and 76, 77, 78, 80-82, 87-89, 109, 114, 116-123, 129, 157, 159, 161, 162, 164, 165, 166, 167, 169-175, 177-183, 223, 224. 232, and 233 above, and further in view of Devine et al (US 5,677,170, 10-14-1997) and Joshi et al.

The teachings of Treco et al and Joshi have been summarized previously (paragraphs 19- 21). None of these teach to include transposition signal in their vectors.

Joshi teaches that the use of transposons in vectors is a known method of cloning new bacterial genes by producing bacterial mutants by random integration of plasmids containing transposons and then identifying and cloning the mutated genes (see last paragraph in column 2 on page 242).

Devine et al teach efficient methods of creating artificial transposons and inserting these transposons into plasmid targets in vitro. These can be used to introduce any functional or non-functional DNA cis elements, sequence or combination into another segment of DNA (see lines 11-67 col 10 continued in cols 11 and 12). They also teach that in their experiments the transposon integration withing targets was sufficiently random that insertion could be recovered

from all regions of target plasmids (see lines 17-19, column 6). Additionally, Devine et al disclose that transposons can be used for mapping genes, chromosome fragments, promoters, enhancers, introns and exons (see columns 9-11).

At the time of the invention, it would have been obvious to an artisan of ordinary skill in the art to modify the vector of Treco et al by including transposition sequences of Devine et al with reasonable expectation of success and use such vectors in cloning new genes by the method of Joshi or identify splice sites in DNA fragments or in genomic sequences in a cell. An artisan would have been motivated to include the transposon in the vector because it would result in the integration of the vector at different sites in the genome of a cell due to random integration.

29. Therefore, the claimed invention as a whole would have been *prima facie* to one of ordinary skill in the art at the time the invention was made in the absence of evidence to the contrary.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Ram R. Shukla whose telephone number is (703) 305-1677. The examiner can normally be reached on Monday through Friday from 7:30 am to 4:00 pm.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Karen Hauda, can be reached on (703) 305-6608. The fax phone number for this Group is (703) 308-4242.

Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to the Group receptionist whose telephone number is (703) 305-0196.

Ram R. Shukla, Ph.D.

SCOTT D. PRIEBE, PH.D
PRIMARY EXAMINER